



SEQUENCE LISTING

<110> Hannoufa, Abdelali
Hegedua, Dwayne
Bate, Nicholas

<120> A Repressor-Mediated Regulation System for Control of Gene Expression
in Plants

<130> 1096.021A

<140> US 10/719,996
<141> 2003-11-21

<150> PCT/CA02/00740

<151> 2002-05-23

<150> US 60/292,973

<151> 2001-05-23

<160> 44

<170> PatentIn version 3.0

<210> 1

<211> 429

<212> DNA

<213> Agrobacterium tumefaciens

<400> 1
atgacggaaa ctgcatacgg taacgcccag gatctgctgg tcgaactgac ggccggatatt 60
gtggctgcct atgttagcaa ccacgtcggtt ccggtaactg agcttcccggt ccttatttcg 120
gatgttcata cggcactcag cggAACATCG gcACCCGGcat cggTGGCGGT caatgttcaa 180
aAGCAGAAGC ctgctgtgtc ggTCGCAAG tcggTTcagg acgatcatat cgtctgtttg 240
gaatgtggtg gtcgTTcaa gtcgCTCAA CGCCACCTGA CGACGcatca cAGCATGACG 300
ccggaagaat atcgcgaaaa atgggatctg ccggTCGATT atccgatgggt tgctcccgcc 360
tatGCCGAAG cccgttgcg gtcgccaag gaaatgggtc tcggTCAGCG ccgcaaggcg 420

aaccgttga

429

<210> 2

<211> 458

<212> DNA

<213> Artificial sequence

<220>

<223> synthetic ROS optimized for plant codon usage and encoding fusion of ROS and nuclear localization signal

<400> 2

atgactgaga ctgcttacgg taacgctcag gatcttcttg ttgagcttac tgctgatatc 60

gttgctgctt acgtttctaa ccacgttggt cctgttactg agcttcctgg acttatctct 120

gatgttcata ctgcactttc tggAACATCT gctcctgctt ctgttgctgt taacgtttag 180

aagcagaagc ctgctgttgc tggtcgtaag tctgttcagg atgatcatat cgtttgg 240

gagtgtggtg gttcttcaa gtctctcaag cgtcaccctta ctactcatca ctctatgact 300

ccagaggagt atagagagaa gtgggatctt cctgttgatt accctatggt tgctcctgct 360

tacgctgagg ctcgttctcg tctcgctaag gagatgggtc tcggtcagcg tcgtaaggct 420

aaccgtccaa aaaagaagcg taaggtctga gagctcgc 458

<210> 3

<211> 447

<212> DNA

<213> Artificial sequence

<220>

<223> ROS consensus sequence

<220>

<221> misc_feature

<222> (1)..(447)

<223> n is A or T or G or C

```
<220>

<221> misc_feature

<222> (1)..(447)

<223> h is a or c or t/u
```

```
<220>

<221> misc_feature

<222> (1)..(447)

<223> m is a or c
```

```
<220>

<221> misc_feature

<222> (1)..(447)

<223> r is G or A
```

```
<220>

<221> misc_feature

<222> (1)..(447)

<223> y is t/u or c
```

```
<220>

<221> misc_feature

<222> (1)..(447)

<223> w is a or t/u
```

<400> 3	
atgacngara cngcntaygg naaygcncar gayytnytng tngarytnac ngcngayath	60
gtngcngcnt aygtwnsnaa ycaygtngtn ccngtnacng arytnccnng nytnathwsn	120
gaygtncaya cngcnytnws nggnacnwsn gcnccngcnw sngtngcngt naaygtngar	180
aarcaraarc cngcngtnws ngtnmgnaar wsngtncarg ayygacayat hgtntgyytn	240
gartgyggng gnwsnttyaa rwsnytnaar mgnacayytna cnacncayca ywsnatgacn	300

ccngargart aymngaraa rtggayytn ccngtngayt ayccnatggt ngcnccngcn	360
taygcngarg cnmgnwsnmg nytngcnaar garatgggny tngncarmg nmgnargcn	420
aaymgnccna araaraarmg naargtn	447
<210> 4	
<211> 27	
<212> DNA	
<213> Artificial sequence	
<220>	
<223> sense primer for amplifying ROS coding region	
<400> 4	
gcggatccga tgacgaaac tgcatac	27
<210> 5	
<211> 25	
<212> DNA	
<213> Artificial sequence	
<220>	
<223> antisense primer for amplifying ROS coding region	
<400> 5	
gcaagcttca acggttcgcc ttgcg	25
<210> 6	
<211> 36	
<212> DNA	
<213> Artificial	
<220>	
<223> sense primer for amplifying tms2 promoter	

<400> 6
tgccggatgca taagcttgct gacattgcta gaaaag 36

<210> 7
<211> 26
<212> DNA
<213> Artificial sequence

<220>
<223> antisense primer for amplifying tms2 promoter
<400> 7
cggggatcct ttcagggcca tttcag 26

<210> 8
<211> 25
<212> DNA
<213> Artificial sequence

<220>
<223> ROS operator sequence
<400> 8
tatatttcaa ttttatttgtatata 25

<210> 9
<211> 109
<212> DNA
<213> Artificial sequence

<220>
<223> ROS-OPDS oligo for p74-315 construct
<400> 9
atctccactg acgtaaggga tgacgcacaa tcccactatc ctgcgaaga cccttcctct 60
atataatata tttcaatttt attgtaatat aacacggggg actctagag 109

<210> 10

<211> 113
<212> DNA
<213> Artificial sequence

<220>
<223> ROS-OPDA oligo for p74-315 construct

<400> 10
gatcctctag agtccccgt gttatattac aataaaattg aaatatatta tatagaggaa 60
gggtcttgcg aaggatagtg ggattgtgcg tcatccotta cgtcagtgga gat 113

<210> 11
<211> 107
<212> DNA
<213> Artificial sequence

<220>
<223> ROS-OPUS oligo for p74-316 construct

<400> 11
atctccactg acgtaaggga tgacgcacaa tctatattc aattttattg taatatacta 60
tataaggaag ttcatttcat ttggagagaa cacggggac tctagag 107

<210> 12
<211> 111
<212> DNA
<213> Artificial sequence

<220>
<223> ROS-OPUA oligo for p74-316 construct

<400> 12
gatcctctag agtccccgt gttctctcca aatgaaatga acttccttat atagtatatt 60
acaataaaat tgaaatatacg attgtgcgtc atcccttacg tcagtggaga t 111

<210> 13

<211> 108
<212> DNA
<213> Artificial sequence

<220>
<223> ROS-OPPS oligo for p74-309 construct
<400> 13
atctccactg acgtaagggg tgacgcacaa tctatattc aattttattg taatatacta 60
tataatataat ttcaatttta ttgtaatata acacggggga ctctagag 108

<210> 14
<211> 112
<212> DNA
<213> Artificial sequence

<220>
<223> ROS-OPPA oligo for p74-309 construct
<400> 14
gatcctctag agtccccgt gttatattac aataaaattg aaatatatta tatagtatat 60
tacaataaaa ttgaaatata gattgtgcgt catcccttac gtcagtggag at 112

<210> 15
<211> 59
<212> DNA
<213> Artificial sequence

<220>
<223> ROS-OP1 oligo for p76-508 construct
<400> 15
gatcctatat ttcaatttta ttgtaatata gctatattc aattttattg taatataat 59

<210> 16
<211> 57
<212> DNA

<213> Artificial sequence

<220>

<223> ROS-OP2 oligo for p76-508 construct

<400> 16

cgattatatt acaataaaat taaaatatacg ctatattaca ataaaattga aatatag

57

<210> 17

<211> 24

<212> DNA

<213> Artificial sequence

<220>

<223> sense primer for amplifying Actin2 promoter

<400> 17

aagcttatgt atgcaagagt cagc

24

<210> 18

<211> 24

<212> DNA

<213> Artificial sequence

<220>

<223> antisense primer for amplifying Actin2 promoter

<400> 18

ttgactagta tcagcctcag ccat

24

<210> 19

<211> 27

<212> DNA

<213> Artificial sequence

<220>

<223> ROS operator sequence in ipt gene

<400> 19
tataattaaa atattaactg tcgcatt

27

<210> 20

<211> 10

<212> DNA

<213> Artificial sequence

<220>

<223> consensus ROS operator sequence

<400> 20
watdhwkmar

10

<210> 21

<211> 104

<212> PRT

<213> Agrobacterium radiobacter

<400> 21

Met Thr Thr Ala Tyr Gly Asn Ala Asp Val Thr Ala Asp Val Ala Ala
1 5 10 15

Tyr Val Ser Asn His Val Val Val Thr Gly Ser Asp Val His Thr Ala
20 25 30

Ser Gly Thr Ser Ala Ala Ser Val Ala Val Asn Val Lys Lys Ala Val
35 40 45

Ser Val Arg Lys Ser Val Asp Asp His Val Cys Cys Gly Gly Ser Lys
50 55 60

Ser Lys Arg His Thr Thr His His Ser Met Thr Tyr Arg Lys Trp Asp
65 70 75 80

Val Asp Tyr Met Val Ala Ala Tyr Ala Ala Arg Ser Arg Ala Lys Met
85 90 95

Gly Gly Arg Arg Lys Ala Asn Arg
100

<210> 22

<211> 138

<212> DNA

<213> Artificial sequence

<220>

<223> p74-315 sequence from EcoRV site to ATG codon of GUS

<400> 22

gatatctcca ctgacgtaag ggatgacgca caatcccact atccttcgca agacccttcc 60

tctatataat atatttcaat ttatttgtaa tataaacacgg gggactctag aggatccccg 120

ggtgtcagt cccttatg 138

<210> 23

<211> 136

<212> DNA

<213> Artificial sequence

<220>

<223> p74-316 sequence from EcoRV site to ATG codon of GUS

<400> 23

gatatctcca ctgacgtaag ggatgacgca caatctatat ttcaatttta ttgtaatata 60

ctatataagg aagttcattt catttggaga gaacacgggg gactctagag gatccccggg 120

tggtcagtcc cttatg 136

<210> 24

<211> 137

<212> DNA

<213> Artificial sequence

<220>

<223> p74-309 sequence from EcoRV site to ATG codon of GUS

<400> 24

gatatctcca ctgacgtaag ggatgacgca caatctatat ttcaatttta ttgtaatata 60

ctatataata tatttcaatt ttatttgtaa ataaacacggg ggactctaga ggatccccgg 120

gtggtcagtc ccttatg 137

<210> 25
<211> 237
<212> DNA
<213> Artificial sequence

<220>

<223> p74-118 sequence from EcoRV site to ATG codon of GUS

<400> 25
gatatctcca ctgacgtaag gcatgacgca caatccact atccttcgca agacccttcc 60
tctatataat atattcaat tttattgtaa tataacacgg gggactctag aggatcctat 120
atttcaattt tattgtataa tagctatatt tcaattttat tgtaatataa tcgatttcga 180
acccgggta ccgaattcct cgagtctaga ggatccccgg gtggtcagtc cctttag 237

<210> 26

<211> 142

<212> PRT

<213> Agrobacterium tumefaciens

<400> 26

Met Thr Glu Thr Ala Tyr Gly Asn Ala Gln Asp Leu Leu Val Glu Leu
1 5 10 15

Thr Ala Asp Ile Val Ala Ala Tyr Val Ser Asn His Val Val Pro Val
20 25 30

Thr Glu Leu Pro Gly Leu Ile Ser Asp Val His Thr Ala Leu Ser Gly
35 40 45

Thr Ser Ala Pro Ala Ser Val Ala Val Asn Val Glu Lys Gln Lys Pro
50 55 60

Ala Val Ser Val Arg Lys Ser Val Gln Asp Asp His Ile Val Cys Leu
65 70 75 80

Glu Cys Gly Gly Ser Phe Lys Ser Leu Lys Arg His Leu Thr Thr His
85 90 95

His Ser Met Thr Pro Glu Glu Tyr Arg Glu Lys Trp Asp Leu Pro Val
100 105 110

Asp Tyr Pro Met Val Ala Pro Ala Tyr Ala Glu Ala Arg Ser Arg Leu
115 120 125

Ala Lys Glu Met Gly Leu Gly Gln Arg Arg Lys Ala Asn Arg
 130 135 140
 <210> 27
 <211> 149
 <212> PRT
 <213> Artificial sequence

 <220>
 <223> synthetic-ROS fused to nuclear localization signal
 <400> 27

 Met Thr Glu Thr Ala Tyr Gly Asn Ala Gln Asp Leu Leu Val Glu Leu
 1 5 10 15

 Thr Ala Asp Ile Val Ala Ala Tyr Val Ser Asn His Val Val Pro Val
 20 25 30

 Thr Glu Leu Pro Gly Leu Ile Ser Asp Val His Thr Ala Leu Ser Gly
 35 40 45

 Thr Ser Ala Pro Ala Ser Val Ala Val Asn Val Glu Lys Gln Lys Pro
 50 55 60

 Ala Val Ser Val Arg Lys Ser Val Gln Asp Asp His Ile Val Cys Leu
 65 70 75 80

 Glu Cys Gly Gly Ser Phe Lys Ser Leu Lys Arg His Leu Thr Thr His
 85 90 95

 His Ser Met Thr Pro Glu Glu Tyr Arg Glu Lys Trp Asp Leu Pro Val
 100 105 110

 Asp Tyr Pro Met Val Ala Pro Ala Tyr Ala Glu Ala Arg Ser Arg Leu
 115 120 125

 Ala Lys Glu Met Gly Leu Gly Gln Arg Arg Lys Ala Asn Arg Pro Lys
 130 135 140

 Lys Lys Arg Lys Val
 145

 <210> 28
 <211> 143
 <212> PRT
 <213> rhizobium elti

 <400> 28

Met Thr Asp Met Ala Thr Gly Asn Ala Pro Glu Leu Leu Val Glu Leu
 1 5 10 15
 Thr Ala Asp Ile Val Ala Ala Tyr Val Ser Asn His Val Val Pro Val
 20 25 30
 Ser Asp Leu Ala Asn Leu Ile Ser Asp Val His Ser Ala Leu Ser Asn
 35 40 45
 Thr Ser Val Pro Gln Pro Ala Ala Ala Val Val Glu Lys Gln Lys Pro
 50 55 60
 Ala Val Ser Val Arg Lys Ser Val Gln Asp Glu Gln Ile Thr Cys Leu
 65 70 75 80
 Glu Cys Gly Gly Asn Phe Lys Ser Leu Lys Arg His Leu Met Thr His
 85 90 95
 His Ser Leu Ser Pro Glu Glu Tyr Arg Glu Lys Trp Asp Leu Pro Thr
 100 105 110
 Asp Tyr Pro Met Val Ala Pro Ala Tyr Ala Glu Ala Arg Ser Arg Leu
 115 120 125
 Ala Lys Glu Met Gly Leu Gly Gln Arg Arg Lys Arg Gly Arg Gly
 130 135 140
 <210> 29
 <211> 142
 <212> PRT
 <213> agrobacterium radiobacter

<400> 29

Met Thr Glu Thr Ala Tyr Gly Asn Ala Gln Asp Leu Leu Val Glu Leu
 1 5 10 15
 Thr Ala Asp Ile Val Ala Ala Tyr Val Ser Asn His Val Val Pro Val
 20 25 30
 Thr Glu Leu Pro Gly Leu Ile Ser Asp Val His Thr Ala Leu Ser Gly
 35 40 45
 Thr Ser Ala Pro Ala Ser Val Ala Val Asn Val Glu Lys Gln Lys Pro
 50 55 60
 Ala Val Ser Val Arg Lys Ser Val Gln Asp Asp His Ile Val Cys Leu
 65 70 75 80
 Glu Cys Gly Gly Ser Phe Lys Ser Leu Lys Arg His Leu Thr Thr His
 85 90 95
 His Ser Met Thr Pro Glu Glu Tyr Arg Glu Lys Trp Asp Leu Gln Val
 100 105 110
 Asp Tyr Pro Met Val Ala Pro Ala Tyr Ala Glu Ala Arg Ser Arg Leu

115

120

125

Ala Lys Glu Met Gly Leu Gly Gln Arg Arg Lys Ala Asn Arg
130 135 140

<210> 30

<211> 143

<212> PRT

<213> *rhizobium meliloti*

<400> 30

Met Thr Glu Thr Ser Leu Gly Thr Ser Asn Glu Leu Leu Val Glu Leu
1 5 10 15

Thr Ala Glu Ile Val Ala Ala Tyr Val Ser Asn His Val Val Pro Val
20 25 30

Ala Glu Leu Pro Thr Leu Ile Ala Asp Val His Ser Ala Leu Asn Asn
35 40 45

Thr Thr Ala Pro Ala Pro Val Val Val Pro Val Glu Lys Pro Lys Pro
50 55 60

Ala Val Ser Val Arg Lys Ser Val Gln Asp Asp Gln Ile Thr Cys Leu
65 70 75 80

Glu Cys Gly Gly Thr Phe Lys Ser Leu Lys Arg His Leu Met Thr His
85 90 95

His Asn Leu Ser Pro Glu Glu Tyr Arg Asp Lys Trp Asp Leu Pro Ala
100 105 110

Asp Tyr Pro Met Val Ala Pro Ala Tyr Ala Glu Ala Arg Ser Arg Leu
115 120 125

Ala Lys Glu Met Gly Leu Gly Gln Arg Arg Lys Arg Arg Gly Lys
130 135 140

<210> 31

<211> 16

<212> PRT

<213> *Arabidopsis* sp.

<400> 31

Arg Ile Glu Asn Thr Thr Asn Arg Gln Val Thr Phe Cys Lys Arg Arg
1 5 10 15

<210> 32

<211> 18
<212> PRT
<213> Nicotiana sp.

<400> 32

Arg Arg Leu Ala Gln Asn Arg Glu Ala Ala Arg Lys Ser Arg Leu Arg
1 5 10 15

Lys Lys

<210> 33

<211> 21

<212> PRT

<213> Nicotiana sp.

<400> 33

Lys Lys Arg Ala Arg Leu Val Arg Asn Arg Glu Ser Ala Gln Leu Ser
1 5 10 15

Arg Gln Arg Lys Lys
20

<210> 34

<211> 18

<212> PRT

<213> Zea mays

<400> 34

Arg Lys Arg Lys Glu Ser Asn Arg Glu Ser Ala Arg Arg Ser Arg Tyr
1 5 10 15

Arg Lys

<210> 35

<211> 47

<212> PRT

<213> Unknown

<220>

<223> potyvirus nuclear localization signal

<220>

<221> misc_feature

<222> (12)..(44)

<223> unknown or other amino acid

<400> 35

Lys Lys Asn Gln Lys His Lys Leu Lys Ala Ala Met Xaa Xaa Xaa Xaa
1 5 10 15

Xaa
20 25 30

Xaa Lys Arg Lys
35 40 45

<210> 36

<211> 17

<212> PRT

<213> Xenopus sp.

<400> 36

Lys Arg Pro Ala Ala Thr Lys Lys Ala Gly Gln Ala Lys Lys Lys Lys
1 5 10 15

Leu

<210> 37

<211> 17

<212> PRT

<213> Xenopus sp.

<400> 37

Lys Arg Ile Ala Pro Asp Ser Ala Ser Lys Val Pro Arg Lys Lys Thr
1 5 10 15

Arg

<210> 38
<211> 17
<212> PRT
<213> *Xenopus* sp.

<400> 38

Lys Arg Lys Thr Glu Glu Glu Ser Pro Leu Lys Asp Lys Asp Ala Lys
1 5 10 15

Lys

<210> 39
<211> 17
<212> PRT
<213> *Mus* sp./*Rattus* sp.

<400> 39

Arg Lys Cys Leu Gln Ala Gly Met Asn Leu Glu Ala Arg Lys Thr Lys
1 5 10 15

Lys

<210> 40
<211> 17
<212> PRT
<213> *Homo sapiens*

<400> 40

Arg Lys Cys Leu Gln Ala Gly Met Asn Leu Glu Ala Arg Lys Thr Lys
1 5 10 15

Lys

<210> 41
<211> 17
<212> PRT

<213> Homo sapiens

<400> 41

Arg Lys Cys Leu Gln Ala Gly Met Asn Leu Glu Ala Arg Lys Thr Lys
1 5 10 15

Lys

<210> 42

<211> 17

<212> PRT

<213> Gallus sp.

<400> 42

Arg Lys Cys Cys Gln Ala Gly Met Val Leu Gly Gly Arg Lys Phe Lys
1 5 10 15

Lys

<210> 43

<211> 17

<212> PRT

<213> Homo sapiens

<400> 43

Arg Lys Cys Tyr Glu Ala Gly Met Thr Leu Gly Ala Arg Lys Leu Lys
1 5 10 15

Lys

<210> 44

<211> 17

<212> PRT

<213> Gallus sp.

<400> 44

Arg Arg Cys Phe Glu Val Arg Val Cys Ala Cys Pro Gly Arg Asp Arg
1 5 10 15

Lys